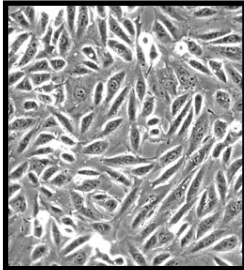


## RENAL SYSTEM INNOPROFILE™ RAT RENAL TUBULAR EPITHELIAL CELLS



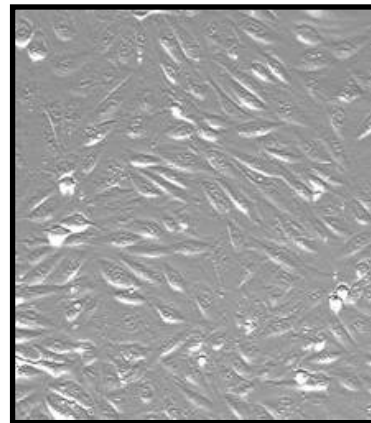
<b>Product Type:</b>	Cryo-preserved Renal Tubular Epithelial Cells
<b>Catalog Number:</b>	P10607
<b>Source:</b>	Rat Kidney ( <i>Sprague Dawley</i> )
<b>Number of Cells:</b>	$5 \times 10^5$ Tubular Epithelial Cells / 1ml
<b>Storage:</b>	Liquid Nitrogen

Rat Renal Tubular Epithelial Cells (RRTEpiC) are isolated from rat renal tissue. Rat Tubular Epithelial cells are cryopreserved at passage one and delivered frozen. RRTEpiC are guaranteed to further expand for 15 population doublings.

Renal tubular epithelial cells (PTEpiC) play a crucial role in renal function. They reabsorb nearly all of the glucose and amino acids in the glomerular filtrate, while allowing other substances of no nutritional value to be excreted in the urine. They are also a major site of injury in a variety of congenital, metabolic, and inflammatory diseases. To be able to study the relationship between tubular cells and a variety of renal diseases, the RTEpiC culture provides a useful in vitro model.

### Recommended Medium

- DMEM/ F-12 HAM  
10ng/ml EGF  
5 µg/ml transferrin  
5 µg/ml insulin  
0.05 µM hydrocortisone



### Product Characterization

Immunofluorescent method

- Cytokeratin-18
- Cytokeratin-19
- Vimentin

The cells test negative for HIV-1, HBV, HCV, mycoplasma, bacteria, yeast and fungi

### Product Use

THESE PRODUCTS ARE FOR RESEARCH USE ONLY. Not approved for human or veterinary use, for application to humans or animals, or for use in vitro diagnostic or clinical procedures

## INSTRUCTIONS FOR CULTURING CELLS

**IMPORTANT:** Cryopreserved cells are very delicate. Thaw the vial in a 37 °C waterbath and return them to culture as quickly as possible with minimal handling!

### Unpacking:

1. For cryopreserved cells: If there is dry ice in the package and you are not going to culture cells right way, place cryovial(s) immediately into liquid nitrogen. If there is no dry ice left in the package, thaw and culture the cells immediately.

### Set up culture after receiving the order:

1. Place the vial in a 37°C waterbath, hold and rotate the vial gently until the contents are completely thawed. Remove the vial from the waterbath immediately, wipe it dry, and transfer it to a sterile field. Rinse the vial with 70% ethanol, and then wipe to remove excess. Remove the cap, being careful not to touch the interior threads with fingers. Using 1 ml eppendorf pipet gently resuspends the contents of the vial.
2. Dispense the contents of the vial into the equilibrated, poly-L-lysine coated culture vessels. A seeding density of 7,500 cells/cm<sup>2</sup> is recommended.
3. Note: Dilution and centrifugation of cells after thawing are not recommended since these actions are more harmful to the cells than the effect of DMSO residue in the culture. It is also important that RTEpiC are plated in poly-L-lysine coated culture vessels that promote the cell attachment growth.
4. Replace the cap or cover, and gently rock the vessel to distribute the cells evenly. Loosen caps if necessary to permit gas exchange.
5. Return the culture vessels to the incubator.

6. For best result, do not disturb the culture for at least 16 hours after the culture has been initiated. Change the growth medium the next day to remove the residual DMSO and unattached cells, then every other day thereafter. A health culture will display polygonal, cobblestone shaped, sheets of contiguous cells and the cell number will be doubled after two to three days in culture.

### Maintenance of Culture:

1. Change the medium to fresh supplemented medium the next morning after establishing a culture from cryopreserved cells. For subsequent subcultures, change medium 48 hours after establishing the subculture.
2. Change the medium every other day thereafter, until the culture is approximately 50% confluent.
3. Once the culture reaches 50% confluence, change medium every day until the culture is approximately 80% confluent

### Subcultures:

1. Subculture the cells when they are 80% confluent.
2. Prepare poly-L-lysine coated cell culture flasks.
3. Warm medium, trypsin/EDTA solution, trypsin neutralization solution, and DPBS to room temperature. We do not recommend warming the reagents and medium at 37°C waterbath prior to use.

4. Rinse the cells with DPBS.
5. Incubate cells with 3 ml of trypsin/EDTA solution (in the case of T-25 flask) until 80% of cells are rounded up (monitored with microscope). Add 3 ml of trypsin neutralization solution to the digestion immediately and gently rock the culture vessel.
6. Harvest and transfer released cells into a 15 ml centrifuge tube. Rinse the flask with another 3 ml of growth medium to collect the residue cells. Examine the flask under microscope to make sure the harvesting is successful by looking at the number of cells left behind. There should be less than 5%.
7. Centrifuge the harvested cell suspension at 1000 rpm for 5 min and resuspend cells in growth medium.
8. Count cells and plate them in a new, poly-L-lysine coated flask with cell density as recommended.

**Caution:** Handling animal derived products is potentially biohazardous. Although each cell strain testes negative for microbial, diagnostic tests are not necessarily 100% accurate, therefore, proper precautions must be taken to avoid inadvertent exposure. Always wear gloves and safety glasses when working these materials. Never mouth pipette. We recommend following the universal procedures for handling products of human origin as the minimum precaution against contamination [1].

- [1]. Grizzle, W. E., and Polt, S. S. (1988) Guidelines to avoid personal contamination by infective agents in research laboratories that use human tissues. *J Tissue Culture Methods*. 11(4).

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